

Complete Genome Sequence of a Novel Avian Paramyxovirus

François-Xavier Briand, Aurélie Henry, Pascale Massin, and Véronique Jestin

Anses, Ploufragan/Plouzané Laboratory, Ploufragan, France

We report here the complete genome of a new avian paramyxovirus (APMV-11) isolated from common snipes. Sequence data from this virus showed that it has the largest genome of APMV and unusual P gene mRNA editing.

paramyxoviruses are enveloped viruses with a negative singlestranded RNA genome of 13 to 19 kb. This family is subdivided into two subfamilies, Paramyxovirinae and Pneumovirinae. The Paramyxovirinae subfamily comprises five genera: Rubulavirus, Respirovirus, Morbillivirus, Henipavirus, and Avulavirus (5). The genus Avulavirus comprises avian paramyxoviruses (APMV) isolated from avian species. To date, only 10 different APMV serotypes (APMV-1 to -10) have been described (7) based on hemagglutination inhibition assays (3) and confirmed by phylogenetic analysis (1). APMV-2, APMV-3, APMV-6, and APMV-7 may induce respiratory disease and/or a drop in egg production in turkeys; APMV-3 may cause encephalitis in several psittacine species; and APMV-5 is associated with diarrhea and high mortality in budgerigars (1). Finally, Newcastle disease induced by APMV-1 is one of the most serious diseases in poultry, and virulent APMV-1 outbreaks must be reported to the World Organisation for Animal Health (8). During avian influenza (AI) active surveillance in 2010, a viral hemagglutinant agent was isolated in 9-day-old embryonated specific-pathogen-free chicken eggs from cloacal swabs collected from live common snipe (Gallinago gallinago) but was not an AI virus. The virus was negative by a hemagglutination inhibition assay using reference antisera against APMV-1 to APMV-10 (except APMV-5, which was not available). Viral RNA was extracted from allantoic fluid and reverse transcribed to cDNA by the use of Superscript II (Invitrogen) and random hexamers. The complete genome sequence corresponding to a new APMV was obtained using overlapping PCR and Platinum Tag DNA Polymerase (high fidelity; Invitrogen). PCR products were sequenced with an Applied Biosystems 3130 Sanger-based genetic analyzer. A contig containing high-quality trace files was assembled using vNTI (Invitrogen). Genome extremities were acquired using rapid amplification of cDNA ends (RACE) and recircularization strategies (6, 9). The genome was 17,412 nucleotides (nt) long with a GC content of 39%, complying with the rule-of-six of Paramyxovirus (4). To date, this is the largest APMV genome reported. Genome organization was typical of APMV (with the exception of APMV-6), with six genes (3'-NP-P/V/W-M-F-HN-L-5') encoding 8 different proteins. Theoretical amino acid lengths of the eight putative proteins are as follows: NP, 455 amino acids (aa); V, W, and P, 277 aa, 181 aa, and 447 aa, respectively; M, 371 aa; F, 562 aa; HN, 583 aa, and L, 2,251 aa. The highest genomic nucleotide identity (48.9%) was obtained with APMV2/Chicken/ California/Yucaipa/56. P gene mRNAs of paramyxovirus are cotranscriptionally edited in response to a *cis*-acting signal (4). The various editing groups are arranged according to the pattern of G insertion into P gene mRNA editing signals to produce proteins P, V, and W (2). All APMV already described require a +1 or +2 frameshift from P mRNA to obtain the V or W protein, re-

spectively. In contrast, sequence data from this new APMV suggested that editing in the P gene is the same as is seen with mumps virus or simian virus 5. For these viruses, the P gene encodes V mRNA, and the addition of one or two nontemplated G residues in the editing site produces W or P mRNA, respectively. Based on the phylogenetic analysis, we have proposed the Common_snipe/France/100212/2010 isolate as the prototype for a new APMV type, APMV-11.

Nucleotide sequence accession number. The genome of APMV-11 has been submitted to GenBank (accession number JQ886184).

ACKNOWLEDGMENTS

This work was supported by EPIZONE (the Network of Excellence for Epizootic Disease Diagnosis and Control; contract FP6-2004-Food-3-A). We are grateful to C. Allée for excellent technical assistance and P. Brown for correction of the manuscript.

REFERENCES

- 1. Alexander DJ, Senne DA. 2008. Newcastle disease, other avian paramyxovirus, and pneumovirus infections, p 75–116. *In* Saif YM, Fadly AM, Glisson GR, McDougald LR, Nolan LK, Swayne DE (ed), Diseases of poultry, 12th ed. Iowa State University Press, Iowa City, IA.
- Hausmann S, Jacques JP, Kolakofsky D. 1996. Paramyxovirus RNA editing and the requirement for hexamer genome length. RNA 2:1033–1045.
- 3. Kim SH, Subbiah M, Samuel AS, Collins PL, Samal SK. 2011. Roles of the fusion and hemagglutinin-neuraminidase proteins in replication, tropism, and pathogenicity of avian paramyxoviruses. J. Virol. 85:8582–8596.
- Kolakofsky D, Roux L, Garcin D, Ruigrok RW. 2005. Paramyxovirus mRNA editing, the "rule of six" and error catastrophe: a hypothesis. J. Gen. Virol. 86:1869–1877.
- Lamb RA, et al. 2005. Family Paeamyxoviridae, p 655–668. In Fauquet CM, Mayo MA, Maniloff J, Desselberger U, Ball LA (ed), Virus taxonomy: classification and nomenclature of viruse. Eighth report of the International Committee for Taxonomy of Viruses. Academic Press, San Diego, CA.
- Li Z, Yu M, Zhang H, Wang HY, Wang LF. 2005. Improved rapid amplification of cDNA ends (RACE) for mapping both the 5' and 3' terminal sequences of paramyxovirus genomes. J. Virol. Methods 130:154–156.
- Miller PJ, et al. 2010. Evidence for a new avian paramyxovirus serotype 10 detected in rockhopper penguins from the Falkland Islands. J. Virol. 84: 11496–11504.
- 8. OIE. 2009. Chapter 2.3.14. Newcastle disease, p 576–589. *In* OIE terrestrial manual 2009: manual of diagnostic tests and vaccines for terrestrial animals. World Organisation for Animal Health, Paris, France.
- 9. Szymkowiak C, et al. 2003. Rapid method for the characterization of 3' and 5' UTRs of influenza viruses. J. Virol. Methods 107:15–20.

Received 17 April 2012 Accepted 26 April 2012

Address correspondence to François-Xavier Briand, françois-xavier.briand@anses.fr.

Copyright © 2012, American Society for Microbiology. All Rights Reserved. doi:10.1128/JVI.00946-12